AMENDMENTS TO THE SPECIFICATION

Please replace the paragraph beginning on page 16, line 11 and ending on page 17, line 2 with the following amended paragraph:

Accordingly, the present invention provides isolated polynucleotides that encode sequences for CD81 gene SEQ ID NO: 46, which is shown to be associated with treatment responsiveness of HCV patients to interferon-α and ribavirin combined therapy. The polynucleotides include polymorphisms associated with responsiveness of interferon-α and ribavirin and are useful as the probes in screening for HCV infected patients suitable for interferon-α and ribavirin combined therapy. The present invention also provides linkage disequilibrium structure of CD81 SEQ ID NO: 46, haplotype information and its use for prediction of potential responders. The present invention further provides methods for detecting polymorphisms in CD81 gene SEQ ID NO: 46 and its surrounding regions, and methods of detecting a propensity to response to the therapy of interferon-α and ribavirin, using the isolated polynucleotides of the present invention.

Please replace the paragraph at page 17, lines 13-15, with the following amended paragraph:

Fig. 1 shows Table 1 of the primers SEQ ID NO: 1-45 for SNP genotyping of CD81 SNPs SEQ ID NO: 46 with FP-TDI method:

Please replace the paragraph at page 18, lines 14-18, with the following amended paragraph:

Fig. 9 shows Table 3 of the association of SNP markers of CD81 gene SEQ ID NO: 46 and its flanking regions with treatment responsiveness of HCV patients to interferon-α and ribavirin combined therapy;

Please replace the paragraph at page 18, lines 19-21, with the following amended paragraph:

Fig. 10 shows Table 4 of the CD81-SNPrs800136 alleles SEQ ID NO: 46 and associated treatment response status;

Please replace the paragraph at page 18, lines 23-25, with the following amended paragraph:

Fig. 11 shows Table 5 of the CD81-SNPrs800137 alleles SEQ ID NO: 46 and associated treatment response status;

Please replace the paragraph at page 19, lines 1-4, with the following amended paragraph:

Fig. 12 shows Tables 6 and 7 of the CD81-SNPrs800334 alleles SEQ ID NO: 46 and genotypes and associated treatment response status:

Please replace the paragraph at page 19, lines 6-8, with the following amended paragraph:

Fig. 13 shows Tables 8 and 9 of the CD81 –SNPpos1989603 alleles SEQ ID NO: 46 and genotypes and associated treatment response status;

Please replace the paragraph at page 19, lines 10-12, with the following amended paragraph:

Fig. 14 shows Tables 10 and 11 of the CD81-SNPrs2522012 alleles SEQ ID NO: 46 and genotypes and associated treatment response status;

Please replace the paragraph at page 19, lines 14-16, with the following amended paragraph:

Fig. 15 shows Tables 12 and 13 of the CD81-SNPrs2522013 alleles SEQ ID NO: 46 and genotypes and associated treatment response status:

Please replace the paragraph at page 19, lines 18-20, with the following amended paragraph:

Fig. 16 shows Tables 14 and 15 of the CD81-SNPrs800335 alleles SEQ ID NO: 46 and genotypes and associated treatment response status;

Please replace the paragraph at page 19, lines 22-25, with the following amended paragraph:

Fig. 17 shows the haplotype blocks that encompass the 5', 3', and the intragenic region of CD 81 gene SEQ ID NO: 46, and SNPs showing to be associated with response to interferon-α and ribavirin combined therapy;

Please replace the beginning on page 20, line 13 and ending on page 21, line 5, with the following amended paragraph:

It is directed to the investigation of the correlation of clinical responsiveness of HCV infected patients to interferon-α and ribavirin combined therapy with genetic polymorphisms in and surrounding the CD81 gene SEQ ID NO: 46. The results indicate that the treatment responsiveness is associated with the host genotype on CD81 gene SEQ ID NO: 46. The distribution of the allele and genotype of several SNPs upstream the CD81 gene SEQ ID NO: 46 is significantly different between the responder and non-responder groups. Further analysis of the linkage disequilibrium structure of the CD81 gene SEQ ID NO: 46 demonstrates that the significant SNPs are clustered in two distinct LD blocks. Moreover, distribution of haplotypes in these two blocks is also significantly different between the responder and non-responder groups. All of these results

indicate that CD81 SEQ ID NO: 46 may directly involve in the treatment response pathway, and its genetic variations play an important role in determining the therapeutic outcome.

Please replace the paragraph at page 21, lines 7-10, with the following amended paragraph:

For reference, Appendix shows the nucleotide sequences of CD81 gene SEQ ID NO: 46 and its 5'-flanking region extended 5K upstream of exon 1 and the 3'-flanking region extended 1 kb downstream of the poly-A tail.

Please replace the paragraph beginning on page 23, line 15 and ending on page 24, line 16 with the following amended paragraph:

The fragment of CD81 gene SEQ ID NO: 46 are amplified by a two-step PCR reaction. The initial amplification step is a multiplex PCR reaction containing 12 different pairs of PCR primers. The reaction mixture consists of 50 ng genomic DNA, 0.1 μM each of 12 pairs of primer, 0.25 mM dNTP mixture, 100 mM KC1, 20 mM Tris-HC1 Ph 8.3, 0.2% Triton X-100, and 5 mM $MgCl_2$, 10U of Vio*Taq* DNA polymerase (VIOGENE) and 0.05U of *pfu* DNA polymerase

(Stratagene) in a total volume of 100 μL reaction. The reaction is performed by a touchdown program with an initial denaturing at 94°C for 4 min, 10 cycles of melting at 94°C for 40 sec, annealing at 72°C with 1°C decrement per cycle for 40 sec, and extending at 72°C for 1 min 30 sec; for the subsequent 25 cycles, the annealing temperature is 62°C with the same conditions for denaturing and extending procedures, and one cycle of final extension at 72°C for 10 min. Amplification is carried out using 2700 PCR machines (ABI) and the amplified products are purified by membrane ultra-filtration with MultiScreen PCR plate (Millipore) according to the manufacture's instruction. In the next step, specific 791 bp of CD81 SEQ ID NO: 46 product is amplified using the purified multiplex product as template is amplified from the simultaneously amplified products in a 78 to 72 touchdown program as described previously in 1 fold of PCR buffer. U.S. Pat. Application No. 10/446,940 is also attached hereto for more detail to perform a two-step PCR reaction with touchdown programs.

Please replace the paragraph beginning on page 26, lines 1-14, with the following amended paragraph:

Four primers are designed for each SNP site, two for PCR amplification of the DNA fragment containing the SNP site and two for TDI reaction. Primer 3 is employed to design the PCR primers. The PCR primers are designed to have a

melting temperature between 54°C to 56°C. The TDI primers are designed by a program, developed originally by Vieux et al., see E. F. Vieux, P.-Y. Kwok, R.D. Miller, Primer design for PCR and sequencing in high-throughput analysis of SNPs. Biotechniques. (2002) Suppl: 28-30, 32., and modified in house by our bioinformatics group, to have melting temperature between 50°C to 55°C and lengths between 20 to 30 bases (about 10,000 Da). Primers SEQ ID NO: 1-45 used for genotyping of SNPs of CD81 SEQ ID NO: 46 in this study are listed in Table 1 of Fig. 1.

Please replace the paragraph beginning on page 29, lines 13-25, with the following amended paragraph:

Figs. 207 show the sequence traces of the responder and non-responder at and around various CD81-SNP alleles SEQ ID NO: 46, respectively. Fig. 2 shows the sequence traces of the responder and non-responder at and around rs800136. Fig. 3 shows the sequence traces of the responder and non-responder at and around rs800137. Fig. 4 shows the sequence traces of the responder and non-responder at and around rs800334. Fig. 5 shows the sequence traces of the responder and non-responder at and around pos1989603. Fig. 6 shows the sequence traces of the responder and non-responder at and around rs8002522012 and rs8002522013.

Fig. 7 shows the sequence traces of the responder and non-responder at and around rs800335.

Please replace the paragraph beginning on page 32, line 3, with the following amended paragraph:

(1) Selected SNPs of CD81 gene SEQ ID NO: 46

Please replace the paragraph beginning on page 32, lines 5-14, with the following amended paragraph:

Over the selected 70 Kb chromosomal region, including CD81 gene SEQ ID NO: 46 and extending 30 Kb each to the 5' and 3' flanking regions, eighteen SNPs and one insertion are identified among HCV infected patients of Chinese population. The positions, sequences, and allele frequencies are summarized in Table 2 of Fig. 8. Among these polymorphic sites, 16 SNPs have minor allele frequency greater than 10% and considered to be informative markers for the association analysis of genetic polymorphism with treatment response.

Please replace the paragraph beginning on page 32, lines 16-18, with the following amended paragraph:

(2) Association of SNP markers of CD81 gene SEQ ID NO: 46 and its flanking regions with treatment responsiveness of HCV patients to interferon-α and ribavirin combined therapy.

Please replace the paragraph beginning on page 33, lines 6-10, with the following amended paragraph:

Distribution of CD81-SNPrs800136 alleles SEQ ID NO: 46 and the status of treatment response are summarized in Table 4 of Fig. 10. the CD81-SNPrs800136T allele SEQ ID NO: 46 is found to be associated with responding status and thus a favorable allele for drug treatment.

Please replace the paragraph beginning on page 33, lines 12-14, with the following amended paragraph:

(4) Association of CD81-SNPrs800137 allele SEQ ID NO: 46 with treatment responsiveness of HCV patients to interferon-α and ribavirin combined therapy.

Please replace the paragraph beginning on page 33, lines 16-20, with the following amended paragraph:

Distribution of CD81-SNPrs800137 alleles SEQ ID NO: 46 and the status of treatment response are summarized in Table 5 of Fig. 11. The CD81-SNPrs800137T allele SEQ ID NO: 46 is found to be associated with responding status and thus a favorable allele for drug treatment.

Please replace the paragraph beginning on page 33, lines 22-24, with the following amended paragraph:

(5) Association of CD81-SNPrs800334 SEQ ID NO: 46 with treatment responsiveness of HCV patients to interferon-α and ribavirin combined therapy.

Please replace the paragraph beginning on page 34, lines 1-6, with the following amended paragraph:

Distribution of CD81-SNPrs800334 alleles SEQ ID NO: 46, genotypes, and the status of treatment response are summarized in Tables 6 and 7 of Fig. 12. The CD81-SNPrs800334 G allele SEQ ID NO: 46 and the GG genotype are found to be associated with responding status and thus a favorable allele for drug treatment.

Please replace the paragraph beginning on page 34, lines 8-10, with the following amended paragraph:

(6) Association of CD81-SNPpos1989603 SEQ ID NO: 46 with treatment responsiveness of HCV patients to interferon-α and ribavirin combined therapy.

Please replace the paragraph beginning on page 34, lines 12-17, with the following amended paragraph:

Distribution of CD81-SNPpos1989603 alleles SEQ ID NO: 46, genotypes, and the status of treatment response are summarized in Tables 8 and 9 of Fig. 13. The CD81-SNPpos1989603 A allele SEQ ID NO: 46 and the AA genotype are

found to be associated with responding status and thus a favorable allele for drug treatment.

Please replace the paragraph beginning on page 34, lines 19-21, with the following amended paragraph:

(7) Association of CD81-SNPrs2522012 SEQ ID NO: 46 with treatment responsiveness of HCV patients to interferon-α and ribavirin combined therapy.

Please replace the paragraph beginning on page 34, line 23 and ending on page 35, line 3, with the following amended paragraph:

Distribution of CD81-SNPrs2522012 alleles SEQ ID NO: 46, genotypes, and the status of treatment response are summarized in Tables 10 and 11 of Fig. 14. The CD81-SNPrs2522012 T allele SEQ ID NO: 46 and the TT genotype are found to be associated with responding status and thus a favorable allele for drug treatment.

Please replace the paragraph beginning on page 35, lines 5-7, with the following amended paragraph:

(8) Association of DC81-SNPrs2522013 SEQ ID NO: 46 with treatment responsiveness of HCV patients to interferon-α and ribavirin combined therapy.

Please replace the paragraph beginning on page 35, lines 9-14, with the following amended paragraph:

Distribution of CD81-SNPrs2522013 alleles SEQ ID NO: 46, genotypes, and the status of treatment response are summarized in Tables 12 and 13 of Fig. 15. The CD81-SNPrs2522013 A allele SEQ ID NO: 46 and the AA genotype are found to be associated with responding status and thus a favorable allele for drug treatment.

Please replace the paragraph beginning on page 35, lines 16-18, with the following amended paragraph:

(9) Association of CD81-SNPrs800335 SEQ ID NO: 46 with treatment responsiveness of HCV patients to interferon-α and ribavirin combined therapy.

Please replace the paragraph beginning on page 35, lines 20-25, with the following amended paragraph:

Distribution of CD81-SNPrs800335 alleles SEQ ID NO: 46, genotypes, and the status of treatment response are summarized in Tables 14 and 15 of Fig. 16. The CD81-SNPrs800335 T allele SEQ ID NO: 46 and the TT genotype are found to be associated with responding status and thus a favorable allele for drug treatment.

Please replace the paragraph beginning on page 36, lines 1-2, with the following amended paragraph:

(10) Linkage disequilibrium (LD) structure of CD81 gene SEQ ID NO: 46.

Please replace the paragraph beginning on page 36, lines 5-9, with the following amended paragraph:

The LD structure of CD81 gene SEQ ID NO: 46 and its flanking regions extended 30 Kb each to the 5' and 3' ends are constructed by genotypes of the

selected 16 informative SNPs. In this 70 Kb chromosomal fragment of DNA, 9 haplotype blocks are identified as depicted in Fig. 17.